Rapid Defense Against the Next-Generation Biothreat

Bioengineered and emerging pathogens represent a significant threat to human health. The best defense against a rapidly expanding pandemic is to isolate the pathogen quickly from biological samples for analysis. The one persistent technology gap in the process of identifying and quantifying the presence of pathogenic agents has been sample handling and preparation that must precede any assay.

Project Goals

The objective of this project is to replace cumbersome, manual techniques with new automated technologies for sample handling and preparation. Specifically, we will use microfluidics with ultrasonic, electrophoretic, and dielelctrophoretic techniques to separate and purify viruses, the most transmissible and infectious agents, from biological and environmental samples.

Relevance to LLNL Mission

By making it possible to rapidly isolate and detect engineered and naturally emerging biothreats, this project contributes to the nation's defense against bioterrorism, which is central to LLNL's homeland security mission. In addition, this project supports LLNL's mission in bioscience to improve human health.

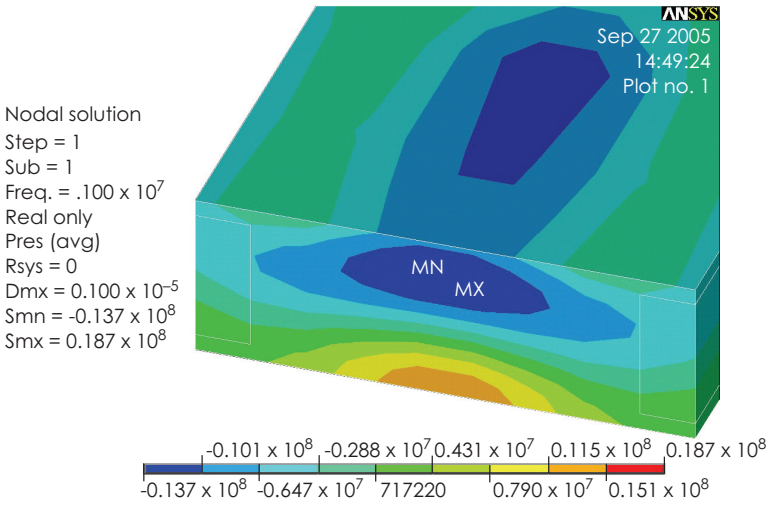

Figure 1. Oblique cross-sectional view of 3-D acoustic model results, showing the predicted shape of an ultrasonic standing wave in a fluid channel. At the chosen frequency, a single stationary node (green region) exists within the channel, as desired, so that particles in the flow will collect at this region for subsequent separation.

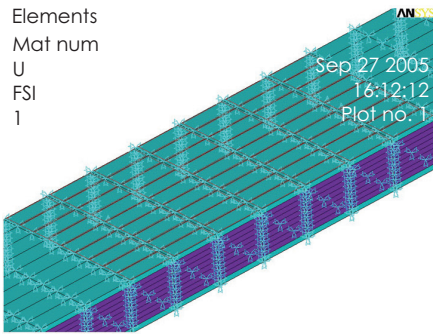

Figure 2. Portion of finite-element acoustic model of a fluid channel, before solving. The top and bottom of the channel are modeled as glass, the sidewalls as plastic (polystyrene).

For more information contact
Raymond P. Mariella, Jr.
(925) 422-8905
mariella1@llnl.gov

FY2005 Accomplishments and Results

In FY2005, we ordered and received the electronics and control instrumentation to set up microfluidic test stations, as well as a selection of fluorescent beads ranging from 30 nm to 5 μm in diameter. These will be used to calibrate our flow system and align the microscope-based test station. Using existing microfluidics models and published data on the physical properties of human cells, bacteria, and viruses, we calculated the scales of systems (channel lengths and cross-sections, electrode size, distributions, and voltages for dielectrophoresis and electrophoresis), to manipulate and separate bacteria and smaller particles from the larger particles.

Figures 1 to 4 show representative results for acoustic/ultrasonic forces.

We expect the new capabilities developed in this project to reduce the time required to identify a new pathogen, such as SARS, by up to an order of magnitude, in part, by better matching methods for sample preparation with the needs of emerging assay technology. These capabilities will also be critical to developing ubiquitous, high-performance autonomous pathogen-sensing systems envisioned as sentinels that monitor for aerosol-transmitted pathogens by screening, for example, air filters or handrails at international airports.

FY2006 Proposed Work

In FY2006, we will instantiate a capability to propagate, characterize, and purify Risk-Group-1 viruses, which will be selected based on the availability of virus and detection reagents. Using simulations that will be continuously validated and improved, we will begin design and fabrication of single-function microfluidic devices to perform manipulations using ultrasonics, electrophoresis, and dielectrophoresis. We will use ultrasonics to disrupt aggregates and divert the unwanted larger particles into the waste stream. Once we have a stream that consists primarily of bacteria and viruses, electrokinetic transport will be used to trap the bacteria and to pass a stream of purified viruses for analysis.

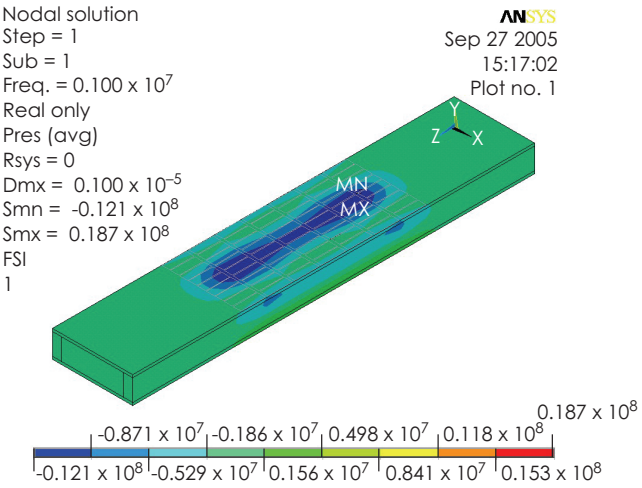

Figure 3. Solved model, showing that the region of the ultrasonic wave stays confined to the area very near the transducers (white outlines), as desired.

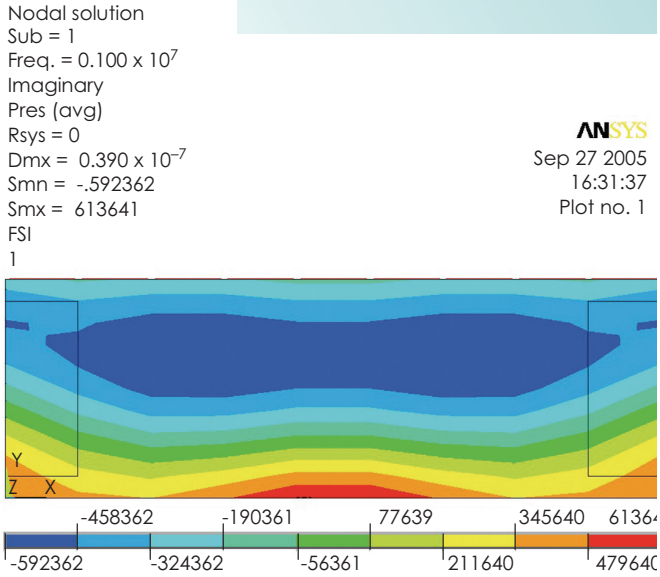

Figure 4. Cross-section of results of a different 3-D acoustic model, showing the detailed shape of an ultrasonic standing wave inside a fluid channel. Waviness in the standing wave is due to the presence of the sidewalls. Finite-element analysis allows for more detailed predictions than those from 1-D theory.